SOME PHYSICAL AND CHEMICAL PROPERTIES OF BONE FROM BOBWHITES

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ABSTRACT. — Some physical and chemical properties of humeri and tarsometatarsi were compared for bobwhites (*Colinus virginianus*) of different sex- and age-classes representing uplands and more fertile bottomlands in southern Illinois. Length, greatest diameter and least diameter of humeri and tarsometatarsi did not differ significantly according to sex, age, sex and age classes, or soil quality. Dry weights of humeri and tarsometatarsi were significantly heavier in males than females, adults than young, young males than young females and adult females than young females. Humeri of birds from the more fertile area were significantly heavier than those from the less fertile area; metatarsi did not differ significantly. Tarsometatarsi generally had greater breaking strengths than humeri. The mean breaking stress of male humeri was significantly greater than those of females; tarsometatarsi did not show significant difference. Humeri of young birds had a significantly greater breaking stress than those of adults; but, tarsometatarsi of adult birds had a significantly greater breaking stress than those of young. Sex and age class comparisons and soil quality comparisons of humeri and tarsometatarsi showed no significant variations. Humeri had a greater percentage of ash than tarsometatarsi in all instances; sex classes showed no significant variation but young birds had greater ash percentages than adults. Among females tarsometatarsi of young had a significantly greater percentage of ash than those of adults. Differences in calcium and phosphorus content of bones from the various sex, age and soil classes were not significant.

In recent years, increasing numbers of ecologists have recognized the importance of soil fertility in its effect on the distribution, abundance and welfare of wild animals. Albrecht (1946) felt that consideration of soil quality must become the basis of wildlife management. Investigations by Leopold and Dalke (1943) with turkeys, Dale (1954) with ring-necked pheasants and Crawford (1950) with rabbits have shown relationships between soil fertility and distribution, productivity and physical and chemical properties of bone.

The purpose of the present investigation was to consider some physical and chemical properties of bones as reflected by sex and age classes of bobwhites and to relate these characteristics to general soil fertility of the collection areas.

METHODS

During January to March, 1956, bobwhite quail were collected in southern Illinois by representatives of the Cooperative Wildlife Research Laboratory. Sex determinations were verified by internal examination. Age was determined by Leopold's (1939) method. Of 195 quail collected, 164 were young and 31 were adults.

Birds were collected from nine southern Illinois counties representing both glaciated and non-glaciated upland soils (Hamilton, Perry, Union, Washington, Jackson, Franklin, Marion and Williamson counties) and Ohio River bottomland soils (Massac County). According to Stefferud (1957), most of the upland soils of southern Illinois are acid and

low in organic matter and plant-food elements. The alluvial soils of the Ohio river bottoms are reasonably fertile showing approximately 4.5 times more organic matter, 3.5 times more nitrogen, 2.6 times more phosphorus, 1.4 times more potassium, 4.1 times more magnesium and 6.2 times more calcium than adjacent upland soils (Wascher et al. 1950). No specific details were available concerning soil improvement practices; however, much of the upland was reported to not have a history of fertilizer usage.

Preparation of Materials

Bones used in all analytical procedures were the left tarsometatarsus and the right humerus. These were chosen on the basis of their similar size and sufficient weight to produce adequate amounts of ash for chemical analyses. If these specific bones were not available in usable condition, their exact opposites were used.

Bones were removed from the birds, fleshed, and cleaned either by hand or dermestid beetles. After cleaning, bones were dried for 2 weeks at 40° C, to equalize moisture content, and then stored in air-tight, glass vials until used in testing procedures.

Physical Determinations

The total length, least and greatest diameters, weight, breaking stress and wall thickness were determined for each tarsometatarsus and humerus. Total length was measured to 0.10 mm with a vernier caliper. Least and greatest diameters at a point midway between the extremities were measured to 0.001 mm with a micrometer caliper. Measurement of the least and greatest diameter was necessary because the bones were not perfectly round; this necessitated the use of both measurements in subsequent calculations of breaking stress. Total weight of the bones was determined on a Roller-Smith balance (calibrated to 0.2 mg).

Breaking stress of an entire bone depends upon strength of the bony material as well as the shape and dimensions of the bone. Breaking stress was determined according to a modification of procedure described by Bell et al. (1941). It was calculated from the formula:

$$S_1 = \frac{M}{a\,b\,t}$$

where S_1 is breaking stress, M is the bending moment at the fracture, a is the greatest diameter, b is the least diameter, and t is the wall thickness at point of fracture.

Wall thickness (t) was measured to 0.1 mm, with an ocular micrometer in a dissecting microscope. Measurements were made on the broken edge of the medial wall of the bone at the point of fracture.

The bending moment (M) is greatest at the midpoint of a long bone (Bell et al. 1941). It was computed from the formula:

$$M = \tfrac{1}{4}\,WL$$

where W is the breaking force and L the length of the bone. In determination of the breaking force (W), the bone was supported at both ends by inserting the ends into depressions in two blocks of wood arranged on a track above table top. A weight pan was suspended by means of a wire hook encircling the exact middle of the bone shaft. Sand was added to a bottle in the pan until the bone broke. Weight of sand, bottle and pan was determined and used as the reading for breaking force.

Chemical Determinations

Chemical determinations were performed on 55 humeri and 61 tarsometatarsi, representing each sex, age and soil class. After breaking, the pieces of the broken bone were weighed and then ashed by a modification of a process described by Morgulis (1931). Ashing was carried out in a muffle furnace at 600° C, for 24 hr. A bone-ash solution was prepared by dissolving 250 mg of the combined ash from a tarsometatarsus and humerus of one bird in 2 ml of concentrated hydrochloric acid and diluted with water to 250 ml in a volumetric flask. Calcium, phosphorus and magnesium were quantitatively determined by analysis of aliquots of this solution.

Calcium was determined volumetrically by a modification of a process described by Furman (1939). It was precipitated from the solution as ammonium oxalate which was dissolved in sulfuric acid and titrated with 0.05 N potassium permanganate.

Phosphorus was measured colorimetrically from an aliquot of the bone-ash solution by the Fiske-Subbarow method as described by Hawk et al. (1947). A Spectronic 20 colorimeter (Bausch and Lomb) was used. In the presentation of data and analyses, all significant differences are based on a 95 percent level of confidence.

Results and Discussion

Physical Determinations

Dimensions: The humeri and tarsometatarsi of all birds had a mean length of 35.5 mm and 32.3 mm, respectively. There were no significant differences according to sex, age, sex and age classes and soil quality. The mean greatest diameters were 3.4 mm for all humeri and 2.6 mm for all tarsometatarsi. There were no significant differences according to sex, age, sex and age classes and soil quality. The mean least diameters of all humeri and tarsometatarsi were 2.6 mm and 2.0 mm, respectively. Least diameters of humeri and tarsometatarsi were not significantly different according to sex, age, and sex and age classes and soil quality.

There were no significant differences in wall thickness according to sex, age, sex and age or soil quality. Values ranged from a low of 0.37 mm for tarsometatarsi of adult males to a high of 0.43 mm for tarsometatarsi of all females.

Weight: The mean dry weight of all humeri was 286.9 mg and 191.2 mg for all tarsometatarsi (Table 1). The humeri of males were heavier than those of females (mean difference 8.1 mg) while the tarsometatarsi of males were heavier than those of females (mean difference 7.0 mg). Humeri and tarsometatarsi of adults were heavier than those of young, the mean difference being 6.3 mg for humeri and 5.6 mg for tarsometatarsi. All of these differences were significant.

Humeri and tarsometatarsi of young males were significantly heavier than those of young females; the mean difference being 9.9 mg between humeri of young males and females and 11.3 mg between tarsometatarsi of young males and females (Table 1). Among adults humeral and tarsometatarsal weights of males were not significantly heavier than those of females. In the case of females humeral and tarsometatarsal weights of young were 12.3 mg and 15.1 mg lighter, respectively, than those of adults; these differences were significant. Humeri and tarsometatari of young males were not significantly different in weight from those of adult males.

Humeri from bottomland area birds were significantly heavier than those from birds of the less fertile upland (mean difference 10.7 mg). A small numerical difference in mean weights of tarsometatarsi (Table 1) between areas of differing fertility was not statistically significant.

In Missouri femurs from rabbits collected on high fertility regions were 12% larger than those from rabbits collected on low fertility regions (Crawford 1950). The lack of correlation in our study with soil fertility may be partially due to the size of the sample or insufficient difference in fertility between upland and bottomland soils. Bell et al. (1941) found that the external dimensions of femora were not altered by varying the calcium intake in the diet of experimental rats.

Breaking Stress: Humeri had a mean breaking stress of 13.4 kg/mm^2, and tarsometatarsi, 16.0 kg/mm^2 (Table 2). The humeri of males had a mean breaking stress 3.2 kg/mm^2 greater than did females. This difference was statistically significant but that for tarsometatarsi was not. One might expect a significant difference

TABLE 1.—Dry weights of humeri and tarsometatarsi of bobwhites, southern Illinois.

	Humerus				Tarsometatarsus			
	Number specimens	Mean weight (mg)	Standard deviation	Range	Number specimens	Mean weight (mg)	Standard deviation	Range
According to Sex								
Female	68	283.0±4.1	33.8	207.8—348.0	72	189.0±3.1	26.4	134.8—316.8
Male	63	291.1±3.5	27.5	250.6—366.8	68	196.0±3.2	26.6	152.0—316.8
Age								
Young	113	286.0±2.3	24.1	207.8—366.8	123	190.5±2.5	27.3	134.8—316.8
Adult	18	292.3±6.7	28.3	235.6—357.6	17	196.1±4.2	17.4	163.6—220.8
Sex and Age Classes								
Young								
female	59	281.3± 3.0	23.0	207.8—348.0	65	185.2±3.4	27.1	134.8—287.6
male	54	291.2± 3.8	27.7	250.6—366.8	58	196.5±3.6	27.6	152.0—316.8
Adult								
female	9	293.6±10.7	32.1	235.6—348.0	7	200.3±6.3	16.7	164.0—220.8
male	9	290.6± 8.9	26.6	264.6—357.6	10	193.1±5.5	17.4	163.6—216.6
Site of Collection								
Massac County	16	294.1±8.6	34.2	244.2—366.8	21	192.2±5.7	26.2	140.4—271.6
All Others	115	283.4±2.3	24.5	207.8—365.6	119	190.8±2.4	26.3	134.8—287.6
Total birds	131	286.9±2.3	26.0	207.8—366.8	140	191.2±2.2	26.3	134.8—316.8

in the breaking stress values of males and females because of the demands of egg laying. We have no valid explanation for the fact that a significant difference was found only for humeri.

Humeri of young birds had a significantly greater breaking stress (2.8 kg/mm^2) than those of adults; however, tarsometatarsi of young had a significantly lesser breaking stress (2.6 kg/mm^2) than adults (Table 2). As one would suspect a significant age variation in breaking stress, this variation seemed contradictory.

TABLE 2.—Breaking stresses of humeri and tarsometatarsi of bobwhites, southern Illinois.

	Humerus				Tarsometatarsus			
	Number specimens	Mean breaking stress (kg/mm²)	Standard deviation	Range	Number specimens	Mean breaking stress (kg/mm²)	Standard deviation	Range
According to Sex								
Female	21	12.2±0.68	3.1	7.2—16.5	21	15.2±0.61	2.8	10.1—20.0
Male	29	15.4±0.56	3.0	8.8—18.9	36	15.6±0.45	2.7	8.7—22.0
Age								
Young	32	15.0±0.51	2.9	7.8—18.0	37	13.7±0.48	2.9	8.7—21.6
Adult	18	12.2±0.73	3.1	7.2—18.0	20	16.3±0.56	2.5	12.4—22.0
Sex and Age Classes								
Young								
female	12	13.1±0.66	2.3	7.8—16.5	12	14.8±0.84	2.9	10.1—20.0
male	20	14.6±0.56	2.5	9.7—18.9	25	15.1±0.60	3.0	8.7—21.6
Adult								
female	9	10.8±0.93	2.8	7.2—14.5	9	15.8±0.37	1.1	12.1—17.5
male	9	13.6±1.03	3.1	8.8—18.0	11	16.7±0.90	3.0	12.4—22.0
Site of Collection								
Massac County	14	13.1±0.86	3.1	7.4—18.9	21	15.3±0.63	2.9	8.7—19.6
All others	36	13.5±0.77	4.6	7.2—18.8	36	15.6±0.45	2.7	10.1—22.0
Total birds	50	13.4±0.36	2.5	7.2—18.9	57	16.0±0.37	2.8	8.7—22.0

The various sex and age class comparisons revealed no significant difference in the mean breaking stress of either the humeri or the tarsometatarsi (Table 2). Although not statistically significant, humeri of adult females had the least breaking stress. These data agree with the data on ash percentages.

There were no significant differences between the breaking stress values of humeri or tarsometatarsi of birds from the bottomland area and those from the upland area (Table

2). Thus, the breaking stress did not vary with the fertility of the southern Illinois soil areas represented in this study. However, Crawford (1950) found that the breaking strength of bones from Missouri rabbits varied directly with the fertility of the soil from which they were collected. Bell et al. (1941) noted that when the calcium intake of rats was increased, the bending moment, a measure of the strength of the whole bone, also increased up to an intake of 36%, after which the bone did not seem stronger. The strength of the bony material, as indicated by breaking stress tests similar to the ones used in our study, was unaffected by the amount of calcium in the diet.

Percent Ash: There were no significant differences in the percentage of ash in the humeri or tarsometatarsi of males and females. But, there was a significant difference between the ash content of young and adults; humeri of the former averaged 1.4% more ash than those of the latter. The ash content of tarsometatarsi of young birds was 3.0% greater than that of adults (Table 3). These values suggest an increased utilization or decreased deposition of inorganic minerals in the bones with increasing age.

Ash content of tarsometatarsi was 4.1% higher for young than adult females (Table 3). The significant difference was expected because of the activity of estrogen in the adult females prior to the egg-laying period. Calcium is mobilized by estrogen from the skeleton and held in the bloodstream for eggshell formation (Nalbandov 1948).

No other significant sex or age difference in the ash content of the bones was found according to sex and age. There were no significant differences in the humeral or tarsometatarsal ash content of birds from bottomland when compared with those of the upland.

Calcium: Comparison of the mean percent calcium of bones representing the various sex, age, sex and age and soil classes revealed no significant variations. The mean percent calcium was approximately 38.0, ranging from 35.2% to 41.1%.

Phosphorus: The percent phosphorus in bones did not differ significantly according to sex, age, sex and age or soil quality. The mean percent phosphorus was approximately 18.6 ranging from 15.8% to 20.0%.

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TABLE 3.—Percentage of ash in humeri and tarsometatarsi of bobwhites, southern Illinois.

	Humerus				Tarsometatarsus			
	Number specimens	Mean percent ash	Standard deviation	Range	Number specimens	Mean percent ash	Standard deviation	Range
According to Sex								
Female	24	65.2±0.37	1.8	62.4—70.0	24	55.5±0.72	3.5	41.7—64.1
Male	31	66.5±0.56	3.1	63.2—72.8	36	57.0±0.65	3.9	47.1—67.3
Age								
Young	35	66.4±0.32	1.9	63.2—72.8	40	57.7±0.57	3.6	41.7—67.3
Adult	20	65.0±0.38	1.7	62.4—67.8	20	54.7±0.89	4.0	46.8—61.3
Sex and Age Classes								
Young								
female	13	65.7±0.50	1.8	63.3—70.0	14	57.4±0.78	2.9	41.7—64.1
male	22	66.8±0.45	2.1	63.2—72.8	25	57.5±0.82	4.1	48.6—67.3
Adult								
female	9	64.6±1.90	5.7	62.4—67.8	11	53.3±1.30	4.3	46.8—61.3
male	11	65.5±1.17	3.9	63.2—67.6	10	55.9±1.76	3.4	47.1—59.2
Site of Collection								
Massac County	16	66.1±0.53	2.1	63.6—72.8	21	55.3±0.68	3.1	46.8—60.1
All others	39	65.8±0.29	1.8	62.4—60.2	39	57.4±0.61	3.8	41.7—64.1
Total birds	55	65.9±0.24	1.8	62.4—72.8	60	56.6±0.49	3.8	41.7—67.3